

Sub
D1

5 1. A synthetic gene encoding a protein normally expressed in an eukaryotic cell wherein at least one non-preferred or less preferred codon in a natural gene encoding said protein has been replaced by a preferred codon encoding the same amino acid, said synthetic gene being capable of expressing said protein at a level which is at least 110% of that expressed by said natural gene in an *in vitro* mammalian cell culture system under identical conditions.

10 2. The synthetic gene of claim 1 wherein said synthetic gene is capable of expressing said protein at a level which is at least 150% of that expressed by said natural gene in an *in vitro* cell culture system under identical conditions.

15 3. The synthetic gene of claim 1 wherein said synthetic gene is capable of expressing said protein at a level which is at least 200% of that expressed by said natural gene in an *in vitro* cell culture system under identical conditions.

20 4. The synthetic gene of claim 1 wherein said synthetic gene is capable of expressing said protein at a level which is at least 500% of that expressed by said natural gene in an *in vitro* cell culture system under identical conditions.

25 5. The synthetic gene of claim 1 wherein said synthetic gene comprises fewer than 5 occurrences of the sequence CG.

6. The synthetic gene of claim 1 wherein at least 10% of the codons in said natural gene are non-preferred codons.

7. The synthetic gene of claim 1 wherein at least 5 50% of the codons in said natural gene are non-preferred codons.

8. The synthetic gene of claim 1 wherein at least 10 50% of the non-preferred codons and less preferred codons present in said natural gene have been replaced by preferred codons.

9. The synthetic gene of claim 1 wherein at least 90% of the non-preferred codons and less preferred codons present in said natural gene have been replaced by preferred codons.

15 10. The synthetic gene of claim 1 wherein said protein is normally expressed by a mammalian cell.

11. The synthetic gene of claim 1 wherein said protein is a retroviral protein.

20 12. The synthetic gene of claim 1 wherein said protein is a lentiviral protein.

13. The synthetic gene of claim 11 wherein said protein is an HIV protein.

25 14. The synthetic gene of claim 13 wherein said protein is selected from the group consisting of gag, pol, and env.

15. The synthetic gene of claim 13 wherein said protein is gp120.

16. The synthetic gene of claim 13 wherein said protein is gp160.

5 17. The synthetic gene of claim 1 wherein said protein is a human protein.

D₂ Sub 18. The synthetic gene of claim 1 wherein said human protein is Factor VIII.

19. The synthetic gene of claim 1 wherein 20% of 10 the codons are preferred codons.

20. The synthetic gene of claim 18 wherein said gene has the coding sequence present in SEQ ID NO:42.

21. The synthetic gene of claim 1 wherein said protein is green fluorescent protein.

15 22. The synthetic gene of claim 20 wherein said synthetic gene is capable of expressing said green fluorescent protein at a level which is at least 200% of that expressed by said natural gene in an *in vitro* mammalian cell culture system under identical conditions.

20 23. The synthetic gene of claim 20 wherein said synthetic gene is capable of expressing said green fluorescent protein at a level which is at least 1000% of that expressed by said natural gene in an *in vitro* mammalian cell culture system under identical conditions.

D [24. The synthetic gene of claim 21 having the sequence depicted in Figure 11 (SEQ ID NO:40).

13 25
25. An expression vector comprising the synthetic gene of claim 1.

5C 16 13 25
26. The expression vector of claim 21, said expression vector being a mammalian expression vector.

Sub D3
10 27. A mammalian cell harboring with the synthetic gene of claim 1.

15 28. A method for preparing a synthetic gene encoding a protein normally expressed by mammalian cells, comprising identifying non-preferred and less-preferred codons in the natural gene encoding said protein and replacing one or more of said non-preferred and less-preferred codons with a preferred codon encoding the same amino acid as the replaced codon.